

CONTRIBUTION TO THE BIOLOGY OF CERTAIN AQUATIC LEPIDOPTERA.*

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Insects of the order Lepidoptera are so pre-eminently terrestrial in organization and habit that the existence of aquatic species is not commonly known, even among biologists. Information concerning American forms which inhabit the water is meager and only a few of their remarkable adaptations have been described. Klapálek and Grünberg ('09, pp. 96-159) discuss sixty species and varieties, which they consider as aquatic, from the fresh waters of Germany and there is reason to believe that ultimately the American aquatic species will be found to exceed that number. During the summers of 1911-15 the writer made some studies of the Lepidoptera of Northern Michigan in connection with which special attention was given to the aquatic group. The data, which form the basis of this paper, were secured in the immediate vicinity of Douglas Lake, in the extreme northern part of the Southern Peninsula. Certain portions of the margins of this lake are rich in aquatic vegetation and afford good breeding grounds for a considerable variety of aquatic animal life. Adults of five species of the genus *Nymphula* (Welch, '15, p. 118) were found in some abundance about the protected bays and beach pools. This genus is unique in being one of the very few groups of Lepidoptera which include species having aquatic stages in the life history. Of the species of *Nymphula* known to occur in North America, six are aquatic in some or all of the immature stages. The larvæ of several other species are not known, but it is probable that they, too, will be found to be aquatic.

In the past, generic divisions have been made in the group of species now included under *Nymphula*, based mainly on certain structural characters of the larvæ. Those species having tracheal gills in the larval stage were assigned to a separate genus, *Paraponyx*, a name still retained by some foreign workers. In this paper, the writer follows the present American practice of including the group assigned to *Paraponyx*, together with others, under the genus *Nymphula*, even though two distinct types of larvæ are thus involved.

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Nymphula maculalis Clemens.

Nymphula maculalis occurred in some abundance in the immediate vicinity of the yellow water-lily beds and the writer was able to observe the development of this species, both in the field and in the aquaria, from the egg to the adult several times during the five successive summers, thus establishing the identity of the following described immature stages.

THE EGG (Pl. VII, Fig. 5).

The eggs were first described by Forbes ('10, p. 221) as follows: "Eggs were laid in captivity, but not in a regular egg-mass. They were oval and flattened, .65 by .5 mm., and had no decided longitudinal ridge. Duration of stage about ten days." This meager description, which seems to be the only one that has been published, contains no hint of the very interesting method of deposition under natural conditions. The writer made a careful study of this part of the life history and the results will be given in some detail.

Place and Method of Deposition.—Eggs were found only on the yellow water-lily, *Nymphaea americana* (Provancher) Miller & Standley. Other aquatic and semi-aquatic plants growing in the vicinity of the yellow water-lily beds were examined carefully but no eggs were found on them. They are laid on the lower side of the floating water-lily leaf and are thus constantly submerged.

Oviposition is associated in a very interesting and unique way with the deposition of the eggs of another aquatic insect. During the past five summers, the writer found about one hundred egg masses of *N. maculalis* in the field and, without exception, all were deposited in the manner to be described. A species of chrysomelid beetle (*Donacia* sp.), which occurs in considerable abundance about the water-lily beds of Douglas Lake, deposits its eggs on the lower surface of the leaves of the yellow water-lily in a very characteristic way. The female cuts a circular hole, 4–6 mm. in diameter, through the leaf and then, by means of a long ovipositor, lays two concentric rows of white, elliptical eggs around the margin on the lower surface. These concentric rows are very rarely complete, so that there may be two or more groups of eggs around the hole with varying intervening spaces. The interesting thing is the fact that the female

of *N. maculalis* uses the holes made by *Donacia* sp. for the deposition of her eggs, thus taking advantage of the work of the female beetle. The eggs of *N. maculalis* are laid in concentric rows on the lower surface of the leaf, about the circular hole and occupy space not utilized by the eggs of *Donacia*. Recently deposited eggs of both beetle and moth were frequently found about the same hole, showing that oviposition by the former is sometimes followed shortly by egg deposition by the latter. Recently deposited eggs of *N. maculalis* were occasionally found about the egg holes of *Donacia* from which the eggs of the latter had disappeared. The number of concentric rows of *N. maculalis* eggs varies slightly but in the larger egg masses there are six. The eggs are so placed that the long axes are radial with reference to the hole about which they occur. The number of eggs in a cluster varies rather widely. Frequently, the size of the cluster is determined by the amount of space unoccupied by the eggs of *Donacia*. In a few instances, the egg mass completely surrounded a *Donacia* egg hole, eggs of the latter being absent. An examination of a large quantity of these egg masses showed that the number of eggs in each varied from twenty-two to two hundred eighty.

The act of oviposition was not observed and exact data on this activity are lacking. However, the necessities make it possible to draw tentative conclusions. The female of *N. maculalis* has no means of making a hole through the leaf and, since the eggs are laid in the above-described manner, she uses the holes provided by some other agency. Even though the writer has failed, in the collections of five summers, to find a single egg mass in the field which was not associated with the egg holes of *Donacia*, it seems improbable that the female *N. maculalis* is restricted to them for egg deposition. In the region examined, *Donacia* is the only animal which makes a small, symmetrical hole through the leaf, a fact which may account for the constant relation between the egg-laying habits of the two species. Similar holes of different origin, if present, might also be used. Since in most of the egg masses there are six concentric rows so arranged that the row most remote from the margin of the hole is four or five millimeters away, the question arises as to the means whereby the female is able to deposit eggs at such a distance from the edge of the hole. An examination of the females of *N. maculalis* shows that the ter-

minal abdominal segment is elongated, cone-shaped, movable, bent ventrad, and bears a long, protrusible ovipositor. Thus it is possible for the female to deposit eggs from the top of the leaf by clinging at the margin of the hole and extending the ovipositor around the edge to the lower surface of the leaf. It also appears that the sixth concentric row of eggs in an egg mass represents the limit of extension of the abdomen and ovipositor.

A number of experiments were performed in order to secure further data on egg deposition. In each experiment, a single female, fresh in appearance and bearing evidences of recent emergence from the pupa, was captured in the field, brought to the laboratory, and placed in a well-lighted aquarium in which a yellow water-lily leaf, bearing no *Nymphula* eggs but having one or more *Donacia* egg holes, was floated on water. Frequent examinations were made of each aquarium during the day and the early part of the night. The results, based upon a number of such experiments, are as follows:

- (1) Deposition of eggs invariably occurred at night.
- (2) Eggs were deposited only about the *Donacia* egg holes.
- (3) A female may deposit eggs for five successive nights before oviposition ceases.
- (4) *Donacia* egg holes, devoid of *Donacia* eggs, were sometimes completely surrounded by six concentric rows of eggs of *N. maculalis*.
- (5) An egg mass, deposited during one night, was sometimes covered, in part, by a second layer of eggs deposited on a succeeding night. Apparently, the deposition of one egg mass on another was an abnormal action, due, possibly, to laboratory conditions, although, aside from the limited space, the conditions in the aquarium resembled those of the natural environment as nearly as possible. Egg masses collected in the field invariably showed but a single layer. It is conceivable that such a reaction might result from the total reduction of available space about the *Donacia* egg holes, but experiments showed that it occurred when the same leaf contained holes which afforded abundant space.
- (6) A single female usually utilized several *Donacia* egg holes before the egg laying period ended.
- (7) The maximum number of eggs laid by a single female was 617.

The method of egg deposition in the absence of *Donacia* egg holes was of interest in this connection. Fresh, vigorous females, captured in the field, were placed in aquaria under conditions simulating the natural ones as nearly as possible. Each aquarium contained yellow water-lily leaves representing one of the following conditions: (1) entire leaves containing no *Donacia* egg holes; (2) leaves containing artificial holes, varying from narrow slits to circular holes, all of various dimensions; (3) pieces of leaves containing no holes and having smooth-cut

edges; and (4) ragged leaves and fragments of leaves with very irregular edges but no holes of any sort.

The results of these experiments were as follows:

- (1) Most of the females tested with entire, unpunctured leaves delayed oviposition until the second or third night and then laid diminutive egg masses on the lower surface of the leaf near the margin, particularly in the region of the leaf sinus.
- (2) Egg masses were deposited around artificial incisions and punctures of various kinds and shapes.
- (3) Apparently, the dimensions of artificial punctures and incisions played little or no part in determining the location of egg masses.
- (4) In the absence of leaf punctures, egg masses were deposited near the edges of smooth-cut portions of leaves, on the lower surface; also about the irregular edges of ragged and mutilated leaves.

It is evident from field observations and the above-described experiments that the female of *N. maculalis* has a distinct preference for the egg holes of *Donacia* in oviposition and that when present they are constantly utilized. However, in spite of the fact that, in the Douglas Lake region, the eggs are normally and constantly placed as above described, it is evident from the experiments that the dependence upon the presence of *Donacia* egg holes is not absolute and it seems fair to assume that in case *N. maculalis* occurs in a locality where *Donacia* egg holes or similar leaf punctures are absent, egg masses would be deposited about the leaf margins of the food plant. Observations on the egg-laying habits of *N. maculalis* in other parts of the continent would be of interest in this connection. This moth is widely distributed, having been reported from localities ranging from the Gulf States to Central Canada. Species of *Donacia* are said to occur in all parts of the United States and Canada and, although not all of them lay their eggs around circular holes in water-lily leaves, it appears that at least one of the rather widely distributed species does have this habit. The extent of this egg-laying interrelation of the two groups of insects is a problem for the future.

Description.—The eggs are elliptical, smooth, slightly compressed, and constant in size. When first hatched, the maximum dimensions are 0.396 mm. and 0.549 mm. The development of the egg is accompanied by an increase in size so that just before hatching the dimensions are 0.450 mm. and 0.648 mm. Of the considerable number of measurements made, none varied more than 0.03 mm. and even this variation was exceptional. It will be noticed that measurements of the egg made

previous to the time of hatching agree quite closely with those reported by Forbes ('10, p. 221). Apparently his measurements were made on eggs which were well advanced in development.

Development.—The egg period was found to be about eleven days, a result which agrees closely with that reported by Forbes. When first laid, the eggs are yellowish, having about the same color as the lower surface of the yellow water-lily leaf. They are also slightly translucent. As development goes on they become darker and proportionately more conspicuous on the leaf. During the first 2–4 days following oviposition, little if any change is apparent but subsequently internal differentiation becomes noticeable. The eggs develop uniformly and usually after a lapse of 3–5 days each begins to show a dark band within, shaped somewhat like the letter J. The position of this dark band is constant, the more curved end being in that extremity of the egg remote from the margin of the hole in the leaf. This band gradually becomes more conspicuous, ultimately revealing itself as the developing body of the larva. The triangular, chitinized pieces of the epicranium begin to appear at the end of the fifth day and it was noted that they develop in that end of the dark band nearest the margin of the leaf puncture, showing that there is a definite orientation of the egg and that the head of the larva develops at the less curved end of the band. At the close of the eighth day, the larva is quite distinct and occupies practically all of the space within the egg. At the end of about ten days, the egg has lost all of the original yellow appearance and the shell has become transparent. The tiny caterpillar can be easily examined and it begins to show movements within the egg. The primary regions of the body are now distinct (Fig. 4), the epicranium and the tips of the mouth parts are dark brown, the fronto-clypeal and the occipital regions are light yellow in appearance, and the chitinized, dark brown prothoracic shield and the dark bands in the regions of the intersegmental grooves are distinctly visible through the egg shell. In the eggs studied in this connection, the dark color appeared first in the ocelli and on the dorsal margins of the intersegmental grooves, later in the head and prothoracic shield. The larva is doubled upon itself with the caudal end extending around and beyond the head.

Hatching.—In almost every case, the entire egg mass hatched at about the same time. Occasionally, a few eggs lagged behind but they hatched not later than five hours after the main group of caterpillars had emerged. The first observed manifestations of the hatching process were motions of the larva within the egg, consisting of a series of body contractions and expansions combined with movements of the head. The mandibles were also in active motion. Apparently, the combined action of the mandibles and the body movements were responsible for the breaking of the egg shell. In the egg masses under observation, the movements of the larvæ preliminary to hatching began from four to six hours before the final escape. The larva emerged from one end of the egg, escaped quickly, and assumed an active habit. In egg masses deposited under natural conditions, the number of imperfect eggs was very small, not exceeding three per cent.

The dates of collection of egg masses varied from July 10 to August 20. Possibly, differences of seasons have some influence on the egg-laying period since in 1913 numerous egg masses were found as late as August 20, while during the preceding summer none were found later than July 30.

THE LARVA (Pl. VII, Figs. 1-3).

First Instar.—The first instar has been briefly described by Forbes ('10, p. 221) as follows: "*Stage I* (.) Slightly larger than *N. gyralis*? described below, with proportionately much larger anal setæ, without trace of gills. Head nearly .3 mm.; length of large anal setæ 1 mm." A careful study of this instar has made it possible to extend the description.

The larva, in this instar (Fig. 1), is light yellowish brown in general appearance. The body is elongate, subcylindrical, and tapers very slightly caudad. Body length of newly hatched larvæ varied from 1.26 to 1.5 mm. The maximum diameter is in the region of the prothorax where it is about 0.32 mm. The main divisions of the body and the intersegmental grooves are distinctly marked. Duration of first instar in specimens reared in aquaria, about 7 days.

Head approximately 0.3 mm. in width, smooth, shining, and dark yellowish brown. Epicranial suture distinct. Front, clypeus and labrum light yellow. Labrum emarginate, setose.

Mandibles toothed, tips blackish. Labial palpi distinct, penultimate segment truncate-conic, ultimate segment smaller and cylindrical, not more than twice as long as thick, terminating distad in three short, stout spines. Maxillary palpi minute and inconspicuous, terminating in three minute articles. Antennæ distinct, basal joint truncate-conic, second segment slender, cylindrical, about three times as long as thick; distal extremity with an apical seta nearly twice as long as segment bearing it, and four minute articles, one of which bears a short seta. Ocelli five, arranged in semicircle just caudad of base of each antenna; dark area in connection with each group. One pair of setæ on epicranium near mid-dorsal line; a similar pair near mesal margins of ocelli groups; two strong setæ on each lateral surface of head near ocelli.

Thoracic segments very finely granulose. Cervical shield broad, black in color, strongly chitinized, smooth, widest at mid-dorsal region; margins regular, anterior margin very slightly conate, posterior margin slightly convex; covers greater part of dorsal surface of prothorax. Meso- and metathorax without traces of black, not strongly chitinized; color uniformly yellowish. Legs similar in size and color, each ending in a single, strong curved claw. Each thoracic segment with three pairs of well-developed setæ on lateral and dorso-lateral surfaces.

Cephalic margin of abdominal segments I–VIII bordered with black; IX–X devoid of dark color. Dorsal surface of VII and VIII darker than other segments. Posterior segments becoming narrower; IX narrow. Prolegs on III–VI and X. One pair of somewhat conical, fleshy projections on lateral aspects of each segment; one short, ventral seta and a longer, dorsal seta, latter about 0.27 mm. long, on each projection. Segment IX with four pairs of well-developed setæ on dorsal and lateral surfaces. Segment X with a pair of short setæ and one pair of longer setæ on dorsal surface; three setæ, two short and one longer, on each lateral surface; two setæ on caudal margin; and two very long setæ, 1.06 mm. in length, extending from the latero-caudal angles of posterior margin.

Second Instar.—This instar, as such, has not been described. Forbes ('10, p. 221), in his account of the life history of this species, states that "stage II" was "not seen; and no sign of leaf-mining was noticed." The same writer describes "stage

III", using an interrogation mark to indicate uncertainty, as follows: "A transparent caterpillar, essentially like the full-grown ones. The maximum number of gill-filaments is two, and the anterior suprastigmatal and the last three pedals have but one. Length about 4 mm.; head .6 mm." The writer has reared larvæ through the early stages from eggs laid by females in the aquaria and observed no indication of an instar between the first and the one described below as the second, the latter corresponding rather closely to the description of Forbes for "*Stage III. (?)*".

A rather surprising change (Fig. 2) takes place at the first ecdysis. The general shape of the larva is not changed but new structures appear. Measurements show the body to be about 2.5 mm. long. It is almost transparent, and the color of the material in the digestive tract shows through the body-wall, giving combinations of yellow and green to the general color.

Head about 0.38 mm. wide, otherwise as in the first instar. Thorax with margins of cervical shield black, remainder translucent; prothorax with five pairs of well-developed setæ; mesothorax with one pair lateral, filamentous gills; length of each gill about equal to width of corresponding segment; metathorax with two lateral, filamentous gills on each side, both of similar length and resembling mesothoracic gills. Legs translucent. Meso- and metathorax devoid of dark color. Abdomen without dark markings; two lateral gill filaments on each side of all segments except last two, one filament with length about equal to width of corresponding segment, the other about one-fourth shorter; penultimate segment with one lateral, filamentous gill on each side, about as long as width of segment; ultimate segment devoid of gills. No gills on dorsal surface of body. Last two segments with several pairs of setæ, one pair of which is terminal in position, long, and stout. Otherwise similar to first instar.

The interesting part of the change from the first to the second instar is the initial appearance of the tracheal gills in the latter. In most of the specimens studied, the lateral gills were constant in all respects, but a few showed variation from the typical condition. One specimen showed a small, extra gill on the left side of the ventral surface of the second abdominal segment. Another specimen bore only one gill on the right side of abdominal segments II, III, IV, and the left side of abdominal segments II and VII.

Third Instar.—The conditions under which these larvæ must be reared make it very difficult to follow the ecdyses so that the exact number of instars can be determined. The following described instar is identified in this paper as the third since, in the examination of many specimens of different ages in the field and laboratory, no indication of an instar between it and the second could be found. Larval changes were also followed as accurately as possible in specimens reared from eggs and the same result was obtained. The writer believes that the results warrant the designation of this form of the caterpillar (Fig. 3) as the third instar.

Length, 4–5 mm. General color pale yellowish. Head about 0.44 mm. wide, otherwise very similar to second instar. Thorax with anterior margin of cervical shield dark, remainder translucent; five pairs of setæ on prothorax; mesothorax with one pair of lateral, filamentous gills and one pair of dorsal, filamentous gills; metathorax with three pairs of filamentous gills, one pair laterad, one pair dorsad, and another pair ventrad. First abdominal segment with three pairs of gills similar in shape and position to those of metathorax; segments II and III, with one pair of dorsal, two pairs of lateral, and one pair of ventral gills; abdominal segments IV and V with two pairs of lateral and one pair of dorsal gills; segments VI, VII, and VIII, with two pairs of lateral gills; segment IX with one pair of lateral gills; segment X devoid of gills; segments IX and X with several pairs of setæ, one of which is terminal in position and much longer than others.

It will be noticed that the above description agrees to some extent with that given by Forbes for "*Stage IV*", the chief difference being in the maximum number of gill filaments. Although the writer has never found an instar in which the maximum number of gill filaments was three, he is inclined to believe that the third instar of this paper is the same as "*Stage IV*" described by Forbes.

Later Instars.—Examination of a large series of caterpillars, varying from the third instar to the full-grown state, showed the existence of a number of types, based on gill characteristics, gradating from the former to the latter. Each successive type is characterized by increase over the preceding one in body size and in the number of gill filaments. Whether or not each type

represents an instar has not been determined and the discussion of later instars will be left for a future paper. The full-grown larva has been studied by Forbes ('10, p. 221) and the specimens studied in this connection agree with his description in most regards, assuming that his table of gill numbers represents an average condition and not a constant one. The most striking change in the later larval instars is the remarkable increase in the number of gills. The maximum number of gills per segment increases from two in the second instar to as many as ten per segment in the full-grown larva. The total number of gills increases from twenty-two in the second instar to one hundred in the last one. The number of gill filaments per segment increases from four in the second instar to as many as forty-eight in the mature larva. The total number of gill filaments on the whole body increases from forty to over four hundred. Such a provision of tracheal gills would seem to be adequate for a wide range of aquatic conditions.'

Activities of the Larva.

Locomotion.—The young larvæ are active from the moment of hatching, crawling restlessly about over the egg shells and the adjacent leaf surface. Locomotion on the water-lily leaves consists exclusively of crawling movements. In the younger instars, particularly in the first, this method of locomotion is slow, often appearing awkward and inefficient. Crawling, in the older instars, is more active and vigorous and, under normal conditions, constitutes a comparatively efficient form of locomotion. The efficiency of crawling as a method of locomotion depends upon the character of the supporting surface. On the yellow water-lily leaf, crawling is accomplished with some degree of ease, except when the leaf, bearing larvæ on the upper surface, is submerged, a condition which seems to require extra effort. Crawling on the glass surfaces of the aquaria is very slow and inefficient, consisting of little more than a mere clinging to the glass, and change of position is accomplished with difficulty. Other surfaces of a smooth, firm nature also afford difficulties in crawling.

At no time during the larval period did the writer observe any evidence of an ability to swim. In this connection, a number of experiments were tried with larvæ of all ages but results were always negative. Random, writhing movements were

exhibited when larvæ were submerged apart from a supporting surface but such movements were ineffective so far as change of position was concerned. However, another form of locomotion will be described later in connection with the discussion of case-making which serves as a substitute for true swimming.

Case-making.—A very interesting phenomenon in connection with the life history of *N. maculalis* is the case-making activities of the larva. This habit is common to the genus *Nymphula*, both in foreign and native species. Of the latter, Packard ('84, p. 824) gave a brief account of case-making by what was apparently *N. icciusalis* Wlk. Hart ('95, pp. 167–172, 176–180) described it in *N. (Paraponyx) obscuralis* Grt. and *N. (Hydrocampa) oblitalis* Wlk. Forbes ('10, pp. 220–21) gave a brief description of case-making in *N. maculalis* Clem.

The observations of the writer confirm, in most respects, the brief description of Forbes on case-making in *N. maculalis*. However, a number of additional data have been secured and will be discussed in some detail. As mentioned above, the larvæ are active from the moment of hatching. They emerge from the eggs and wander about restlessly for a time before starting to feed. In some of the aquaria, this period of preliminary wandering lasted for two or three hours. In the laboratory, the young larvæ seemed to show a preference for the submerged lower side of the leaf on which they were reared. The young larva, under laboratory conditions, soon began to make an incision in the leaf which was extended in such a way that ultimately a portion of the leaf, oval or circular in form and about 2 mm. in maximum dimension, was cut out and drawn back on the lower surface of the yellow water-lily leaf so that the larva was enclosed. Sometimes the larva cuts out the upper surface of its compartment, thus making an independent case composed of two similar pieces of leaf tied together by silken threads.

As the larva increases in size, the cases are outgrown and new ones made. With the older larvæ, case-making is a simple and rather rapid process. In constructing a new case, the larva crawls to the lower surface of a leaf and usually begins work near the periphery so that the resulting piece is cut out of the edge. However, an occasional leaf is found in which the piece has been cut out near the midrib. Certain random, preliminary

movements are often performed, consisting chiefly of an apparent testing of the lower epidermis with the mandibles, before the larva settles down to the work of removing the piece. It works by using most of the length of the body as a radius and bending the anterior region as the incision is extended. After the initial incision is made, the head is held into the cut and a little to one side, thus placing the cutting plane of the mandibles at approximately right angles to the surfaces of the leaf. One-third of the complete incision may be made without changing the position of the posterior part of the body. Occasionally, a larva takes advantage of the overlapping margins of contiguous leaves. Since such a space is usually filled with water, the larva crawls into it, begins work, and in due time cuts an elliptical piece from one or both of the leaves.

The older larvæ utilize the excised pieces in making cases in several ways:

- (1) A single piece may be cut out and tied flatwise against the lower side of the leaf.
- (2) Two similar pieces may be cut out, forming a lens-shaped case, which is then tied flatwise to the leaf, or else becomes independent.
- (3) Several pieces may be cut out, tied together into a case, and attached to the lower surface of the leaf.
- (4) Two pieces may be cut out, tied together flatwise, and then attached endwise to the leaf.

The majority of the cases are placed on the lower, submerged surfaces of the floating leaves. Occasionally, cases containing larvæ are found on the dry, upper surfaces, a fact which suggests the possibility that the larvæ are not entirely dependent upon respiration by tracheal gills. It is a common thing to find numerous cases of various sizes attached to both surfaces of the submerged leaves of *Nymphæa americana*. How these larvæ get on the submerged leaves is not definitely known but it is possible that some of them, by wave action or other mechanical means, are dislodged from the floating leaves and sink to the submerged leaves, or else to the bottom from whence they crawl up the petioles to the leaves. The presence of young larvæ in such situations is discussed later.

The chief functions of the case appear to be (1) protection, and (2) support in the water. The protective function plays an important part in the life of the larva. These caterpillars occur in an environment where predaceous enemies are common and obviously the case is a rather efficient protection. The larva shows a very distinct tendency to respond to slight

mechanical stimuli by immediate retraction into the case, a reaction which probably plays an important part in escaping enemies. Under normal conditions, the larva is apparently very shy, never, according to the observations of the writer, emerging completely from the case except in connection with the construction of a new one. At times, the anterior two-thirds of the body is projected from the case but such periods of partial emergence are of short duration, except when engaged in a special form of locomotion to be described later. Larvæ, removed from their cases and placed in aquaria provided with yellow water-lily leaves, very soon begin the construction of a new case.

The second function of the case—that of support in the water—is vitally connected with an interesting form of locomotion. The specific gravity of the older larvæ is greater than that of water and, unless supported, they will sink. The leaf tissue of the yellow water-lily has a specific gravity distinctly less than that of water and the oblong pieces cut out by the larva in case-making are always buoyant enough to easily support it. Therefore, all detached cases float at the surface and no effort on the part of the larva is required to support itself in the medium. Where larvæ are numerous, individuals are often found crawling over the top of the water-lily leaf, carrying the cases with them. To accomplish this form of locomotion, the larva extends the anterior part of the body from the case, uses the true legs as locomotor organs, and holds the case with the prolegs. When detached cases are dropped in the water, the larva performs certain movements which result in a change of position in space. As mentioned before, no evidence of an ability to swim was observed when larvæ were isolated from their cases. However, they do possess a form of locomotion in water in conjunction with their cases. The anterior portion of the body is projected from the case into the water and vigorous, horizontal, side to side motions are executed which result in the propulsion of both larva and case. It does not constitute a very efficient form of locomotion but is effective enough to bring the larva in contact with other water-lily leaves. If the horizontal movements are equal on either side of the long axis of the body, the result is a backward movement approximately in a straight line. If, as is often the case, the strokes are stronger on one side than the other, the result is an irregular rotation.

The effect of case-making upon the food plant is frequently serious. Some of the yellow water-lily beds (*N. americana*) about Douglas Lake are heavily infested at times with the larvæ of *N. maculalis* and suffer greatly (Figs. 12-19). The total effect of the larvæ on the food plant includes the amount of plant tissue consumed as food and the plant tissue utilized in case construction. According to the observations of the writer, the plant suffers much more from the case-making than from the removal of tissue for food. Case construction results in a reduction of the leaf surface which may be extensive enough to leave only the midrib. The writer observed beds of *N. americana*, in August, which, as nearly as could be estimated, had lost 40 per cent. of the total leaf surface by the case-making activities of these larvæ.

Food.—Very young larvæ, reared in shallow aquaria, fed on the lower side of the yellow water-lily leaf, feeding and case-making being accomplished at the same time. The translucency of the body made it possible to observe the first occurrence of green plant tissue in the digestive tract. After the case was made, the tiny larva fed to some extent upon the tissue of the case. However, the normal field habits of the larva in the first instar were not determined. In rearing young larvæ in aquaria, some difficulty was experienced in securing the second instar and in preventing a very high mortality among those which hatched from egg masses. The following circumstantial evidence suggests that possibly the larvæ, after hatching, settle to the bottom and after one or two ecdyses, approach the surface of the water on the petioles of the water-lily:

- (1) Recently hatched larvæ have a tendency, in the aquaria, to drop to the bottom and to wander about.
- (2) The first and second instars were not found on the floating leaves of the yellow water-lily where the egg masses occurred.
- (3) It was frequently observed that in water-lily beds in which the floating leaves had petioles two or more feet long, the *submerged* leaves bore young larvæ (third instar or a little later) while the floating leaves bore only the more advanced larvæ. Some of the submerged leaves had petioles only about one inch long, so that the leaf was practically on the bottom.
- (4) Very rarely did pupæ occur on the submerged leaves.

Larvæ, hatching in very shallow aquaria, ate the yellow water-lily leaf tissue and some developed into the second instar, but it is possible that it was not the normal reaction and that the first instar may be passed on the bottom. In later instars,

feeding was confined largely to adjacent leaf tissue not a part of the case itself. All excrement was voided outside of the case, thus providing for the cleanliness of the interior.

Forbes ('10, p. 220) states that in addition to *Nymphæa*, *Castalia* and *Brasenia* are used by *N. maculalis* in case-making and his description leads one to infer that all of these plants are used as food. The writer has found no evidence of feeding or case construction involving plants other than *N. americana* although *Nymphæa* and *Castalia* intermingle in the same beds. *Brasenia* does not occur in the Douglas Lake region.

Respiration.—Respiration in the first instar is apparently cutaneous. Tracheal gills have not yet appeared and, since the larva is almost constantly submerged, it would appear that it must of necessity utilize the dissolved oxygen of the water by absorption through the body-wall. Larvæ, hatched and reared in the laboratory, almost invariably sought the lower side of the water-lily leaf regardless of whether the leaf was submerged or floating. A study of the various situations in which the larvæ of *N. maculalis* occurred most abundantly showed that the water contained a considerable amount of dissolved oxygen, due to the exposure to the air and the agitation by wind and wave action. It is possible that the oxygen demands of the larva in the first instar are low enough that they can be satisfied by the cutaneous form of respiration and special organs are not demanded.

After the first ecdysis, tracheal gills appear, each gill containing a primary branch from the longitudinal tracheal trunks. The appearance of about fifty gill filaments in the second instar would seem to be ample provision for the increased oxygen demand. The marked increase in the number of gills and in the number of branches of each gill in the later instars has been discussed.

The occasional appearance of larvæ on the upper surface of the leaf seems to indicate an ability to pass at least a limited time out of water. Instances of pupation on the upper surface of the leaf were observed in the field and the time required for a larva to emerge from the water, locate the case, and construct the silken inner covering would seem longer than the individual could survive without some form of oxygen supply. No data were secured on the mode of respiration under these conditions.

Possibly, cutaneous respiration continues to be operative in the later instars.

Dissemination.—The larvæ have several methods of dispersal:

- (1) Since *N. americana* grows in beds in which the leaves are very frequently contiguous or overlapping, the larvæ hatching from a single egg mass may, by crawling, scatter over a number of leaves. The older larvæ are more efficient at crawling and may ultimately get a considerable distance from the original food plant.
- (2) Larvæ may propel themselves in detached cases from one food plant to another as already described.
- (3) Wind, wave action, and currents are sometimes very effective in scattering detached cases containing larvæ.
- (4) Water-lily leaves, broken from the petioles by wave action or loosened by the attacks of certain species of insects, frequently bear the attached cases of *N. maculalis* and such leaves float about from place to place at the will of the waves and currents. This form of dispersal was very common in some of the water-lily beds about Douglas Lake, where the plants were badly affected by the larvæ of *Hydromyza confluens* (Welch, '14a, pp. 139-140) which cause the petiole to break under very slight side to side strains, and by the larvæ of *Bellura melanopyga* (Welch, '14b, p. 104) which sometimes sever the leaf from the petiole near the upper end of the latter.

PUPA (Pl. VIII, Figs. 6-7).

Forbes ('10, p. 222) gives the following brief description of the pupa: "*Pupa* similar in general form to that of *obscuralis*, as described by Dyar, but of the seven ridges near the tip of the abdomen beneath, only the central one remains, and the anal opening is not distinctly Y-shaped. The case for the hind legs varies considerably in length." Measurements of a large number of pupæ showed that there is some variation in size. The length, exclusive of the spike-like setæ on the vertex, varies from 10 mm. to 14.5 mm., average 12.6 mm. The maximum diameter is in the region of the second to the fourth abdominal segments and, in the specimens examined, the average is 3.4 mm., the extremes being 2.0 mm. and 4.0 mm. The color is uniformly light yellow, except in specimens about ready to transform in which the developing colors of the adult begin to show through the pupal integument. Pupæ preserved in alcohol lose the yellowish appearance and become whitish. The body is rather soft, smooth throughout, naked, and semi-opaque. The anterior region tapers slightly cephalad while the posterior region tapers distinctly caudad. The abdomen is bent so that the ventral surface of the body is nearly plane while the dorsal surface is distinctly convex.

The head is smaller than the prothorax. It bears on the vertex two slender, porrect, very slightly dehiscent setæ, reddish brown in color, and approximately 0.21 mm. in length. In the older pupæ, the eyes are dark in color and show through the integument. The antennal cases lie parallel to the second leg cases, extending with the latter around on the ventral surface of the body and approaching the mid-ventral line. In the specimens examined, the length of these antennal cases varied, depending, apparently, upon the age of the pupa. The extent of the variation is from a position opposite the first pair of abdominal spiracles to the posterior tips of the wing cases. They acquire an external segmentation corresponding to the segmentation of the antennæ within and have a distinctly moniliform appearance. From the ventral surface of the head, a long, tapering, double sheath extends caudad along the mid-ventral line of the body to a point approximately opposite the tips of the wing cases. It contains in its cephalic end the developing palpi and proboscis.

The thoracic segments and their associated parts are similar in color and appearance in the newly transformed pupæ. On the ventral surface, the cases for the wings and legs cover the greater part of the first four abdominal segments. The tips of the wing cases extend almost to the caudal margin of the fourth abdominal segment. The ventral sheath, for the hind legs, is prolonged caudad almost to the tip of the abdomen. As stated by Forbes ('10, p. 222), the length of this ventral sheath varies considerably. In the large number of pupæ examined by the writer, the caudal extremity varies in position from the caudal margin of the sixth abdominal segment to the posterior end of the body, a variation represented in extent by the combined width of the three posterior segments of the abdomen. The sheath for the second pair of legs also varies in length. It often extends but little if any beyond the tips of the wing cases, but specimens have been studied in which it extended to the caudal margin of the fifth abdominal segment. The case for the first pair of legs is shorter than the others, the posterior extremity usually reaching a point opposite the spiracles on the third abdominal segment, although this case is likewise subject to some variation in length. Observations on pupæ of different ages lead the writer to believe that the length of the leg cases is

dependent, to some extent at least, upon the degree of development of the quiescent stage, the length increasing as the time of emergence of the adult approaches. All of the appendages of the head and thorax become more and more apparent and it is often possible to pick out the female pupæ by the uniform dark slaty gray appearance of the front wings. The leg cases become increasingly distinct and acquire an external segmentation corresponding to that of the enclosed leg.

The abdomen is widest at its junction with the thorax and tapers gradually caudad. Except at the extreme posterior end of the body, the intersegmental grooves are broad and well-defined. The second, third, and fourth abdominal segments bear each a pair of large, lateral, conspicuous spiracles, all of nearly uniform size, and borne on fleshy, conical tubercles. The slit-like opening is transverse in position and surrounded by an almost circular, dark orange, chitinized peritreme. Separated from the peritreme by a narrow space and at a slightly lower level is a fine, brown, concentric line. The various structural elements of these spiracles vary in size in the different specimens and to a limited extent in the same specimen. The average dimensions of the peritreme are about 0.232×0.264 mm., the average length of the slit-like opening is approximately 0.14 mm., and the average diameter of the outer ring is about 0.28 mm. Vestigial spiracles occur on abdominal segments V-VII but are so inconspicuous that close examination under magnification is usually required to locate them. The ventral surface of the tip of the abdomen bears a number of short, longitudinal carinæ, arranged in two groups. The intersegmental grooves in this region are obscure and careful examination is required to determine the segmental position of these groups of carinæ. The anterior group occurs on the eighth abdominal segment and usually consists of a single, median carina or a pair of similar carinæ, one on either side of the median line. The second group of carinæ occurs near the anterior margin of the ninth abdominal segment and consists of a single, median carina separating the components of a pair of similar carinæ. Sometimes a second pair of lateral carinæ are also present. The crests of the carinæ in both groups are brownish in color while the sides have the same color as the adjacent parts of the abdomen.

The number and arrangement of these carinæ on the ventral surface of the ninth abdominal segment present considerable variation. Forbes ('10, p. 222) states that this pupa is similar in general form to that of *N. obscuralis* "but of the seven ridges near the tip of the abdomen beneath, only the central one remains". The writer has examined a large number of these pupæ during the past five summers and has failed to find a single pupa which possessed only the single, median carina. The first pair of lateral carinæ is always present in addition to the median one and, as stated above, there is sometimes present a second pair laterad of the first. When a second pair is present, the component carinæ are usually smaller than the other carinæ and not so readily distinguished. In *N. obscuralis*, as described by Hart ('95, p. 173), the first group of elevations are described as follows: "Apex of abdomen subacute; ninth segment beneath with a faint elevated line at middle, and a small elevation each side." In the figure accompanying this description, the "small elevation each side" is represented as circular in contour. This condition seems distinctly different from that existing in *N. maculalis* since, in all of the pupæ examined, the single elevated line, when present, is unaccompanied by conical elevations of any sort, and when absent, it is represented by a pair of similar, longitudinal elevations. Furthermore, this group of elevations is on the eighth abdominal segment. The above description probably does not include all of the variations which exist since the writer found one specimen in which the anterior group of carinæ consists of three pairs of elevations, diminishing in size on either side.

Caudad of the second group of carinæ is a distinct Y-shaped impression. Forbes ('10, p. 222) makes the following statement: ". and the anal opening is not distinctly Y-shaped." All of the specimens examined in this connection show this ventral impression to be of such structure that the expression "Y-shaped" describes it quite well.

As already stated, the full-grown larva usually attaches its case to the lower, submerged surface of a water-lily leaf and transforms into the pupa. Just before pupation, a dense, whitish, apparently complete, silken covering is spun around the larva. This silken covering adheres closely to the inner walls of the case and is strongly attached at the periphery. It is

elliptical in outline, the dimensions usually being about 1.25×2 cm. The maximum thickness is usually but little more than the maximum diameter of the pupa. This silken covering is relatively strong and not easily torn open.

The case varies to some extent in size and shape. In general, it is elliptical and has dimensions varying from 1.5×2 cm. to 2.5×5 cm. The vast majority of the pupal cases are attached to the lower surface of the floating water-lily leaves and are thus constantly submerged. Pupal cases have been found on the upper surface of floating water-lily leaves, a position which minimizes contact with the water. The fate of such pupæ is not known. A few pupal cases have also been found on the submerged water-lily leaves, several inches under the surface of the water.

The external surface of pupæ is not easily wetted and they float if removed from the silken coverings and placed in water. The fate of pupæ which accidentally become removed from their cases is not known. It is said (Miall, '95, p. 233) that the pupæ of certain European species of *Nymphula*, when removed from their silken coverings and placed in water, live for a time but do not transform into adults.

Dissemination.—In connection with the discussion of the dissemination of the larvæ, mention was made of the fact that an indirect method of distribution occurs, due to the influence of wind and waves, or the work of other insects, or both. The pupæ are sometimes scattered in the same way. The work of the larvæ of *Hydromyza confluens*, *Bellura melanopyga*, and others, frequently bring about the separation of the leaf or portions of the leaf from the petiole, permitting them to float about at random. Such detached leaves often bear pupal cases of *N. maculalis* and may be carried some distance from the original breeding place. After storms in which portions of the floating water-lily leaves are torn away by wave action, pupæ are sometimes found on the opposite shore.

THE ADULT.

Adults of *N. maculalis* have been taken about Douglas Lake by the writer from June 28 to August 30 but these limits are probably too narrow. They are normally confined to the region surrounding the food plants of the larva and the few scattering adults which are sometimes found remote from yellow water-

lilies are probably individuals which have emerged from pupæ which have been drifted away from the original breeding place by the wind and waves, rather than individuals which have voluntarily wandered from the place of emergence.

The adults of *N. maculalis* are largely nocturnal in habit. Occasionally, individuals may be observed in flight during the day but, in general, the period of activity begins shortly after dusk. During the day, they can usually be driven to flight by walking through the water-lilies or in the vegetation growing near the edge of the water, or by beating the undergrowth fringing the beach near the water-lily zone. On calm, clear days, adults are commonly found at rest upon the upper surface of the water-lily leaves, but on windy days they usually seek the undergrowth near the edge of the beach. They respond readily to slight disturbances by short flights but observations both in the field and in the aquarium lead the writer to believe that voluntary flight during the day is not common. Females in the breeding cages remain inactive throughout the entire day. Diurnal flight is apparently at random and the selection of objects of support a matter of chance. Only one constant feature was observed, namely, adults never made long flights away from the immediate vicinity of the water-lily beds. Individuals, driven from resting positions, often flew over the open water, never rising high above the surface, dropping momentarily on the surface from time to time, but almost invariably, after a flight of three or four rods, returning to the same vicinity. Individuals which happened to fly out from shore during a strong wind had difficulty in returning and were frequently carried far out on the lake.

The adult apparently suffers little if any from contact with water. The usual position of the pupa is such that, on emergence, the adult is, of necessity, compelled to come to the surface through a certain depth of water. Individuals flying over open water, as described above, frequently drop momentarily on the surface, taking wing again without difficulty. Individuals, whose powers of flight have been exhausted, drop ultimately to the surface and may rest there for a considerable length of time.

Nothing was discovered concerning the feeding habits of the adult. Individuals have lived for days in an aquarium without food but the possession of well-developed mouth parts points rather definitely to a feeding habit.

Only incidental observations on the enemies of this species were made in this connection. It is probable that the possession of a case in the larval and pupal stages is a provision whereby considerable protection is afforded. The larvæ occur in situations which are rich in predaceous animals of several kinds and there is no reason to suppose that such fleshy larvæ would be exempt from attack. One of the robber-flies, common in the Douglas Lake region, preys on the adult and specimens of the latter have been taken with adult *N. maculalis* in their claws. Although this robber-fly is a vigorous enemy, it seems probable that the nocturnal habit of the moth prevents greater fatality from this enemy.

***Nymphula icciusalis* Walker.**

Early Stages.

Nymphula icciusalis is abundant on the north shore of Douglas Lake about the bays, beach pools, and inlets, all of which contain quantities of *Potamogeton*, *Vallisneria*, and other aquatic plants. By confining females in aquaria containing the leaves of several aquatic plants, egg masses were easily secured and the process of hatching and the early development of the larva were observed. Since the early stages of the life history have not been described, the following data have been included in this paper. Packard ('84, p. 824) figured a larva, which he supposed to be *N. icciusalis*. Forbes ('10, pp. 225-6) reared a single larva through to maturity and reported close correspondence of data with those of Packard, but made no mention of the eggs or early larval instars. Miller ('12, pp. 127-134, 245) made some observations on this species, described some of the activities of the larvæ, made slight mention of the pupa and of the eggs, and described the full grown larva in considerable detail.

THE EGG.

Place and Method of Deposition.—The following data were taken from egg masses deposited by females in the aquaria and from numerous egg masses taken in the field. Egg masses from both sources were laid on the leaves of *Potamogeton natans*. In aquaria containing only *Nymphæa americana*, females, after some delay, deposited eggs on the leaves of this plant in the characteristic way but this was evidently not a normal reaction.

Eggs are laid in a definite cluster on the lower surface of the leaf and very near the margin. No egg masses were found in connection with punctures or artificial breaks through the leaves. The eggs are arranged in concentric rows, usually four, which are separated by uniform spaces, each row being curved so that the concave side is always towards the margin of the leaf. The number of eggs composing the clusters examined in this connection varies from 17 to 76. Clusters are sometimes placed so closely together that they appear to be almost confluent. The row of eggs nearest the margin of the leaf is always at least 2 mm. from the edge and sometimes as much as 5 mm. Oviposition was not observed but the position of the egg mass and the distance of the rows from the edge of the leaf indicate that possibly the female lays the eggs, in a manner similar to that suggested for *N. maculalis*, by clinging to the edge of the leaf, extending the ovipositor under the edge and swinging it around, thus depositing the eggs in concentric rows.

Description.—The eggs are elliptical in outline, slightly flattened, smooth, uniform in size, and 0.45 mm. x 0.6 mm. in dimensions. They are whitish in color, being distinctly lighter than the leaf surface on which they are placed. When first laid, they show no external signs of internal differentiation.

Development.—The egg period is from ten to eleven days. Eggs, deposited in the aquaria, develop in the same way and at the same rate as the egg masses collected in the field. During the first two days subsequent to oviposition, no internal changes are evident. At the end of about fifty hours, the eggs begin to show signs of internal differentiation and, during the following 24–36 hours, a dark band develops within, similar in some respects to the one which appears in the early development of the eggs of *N. maculalis*. This band has a shape somewhat like the letter J and is constant in its position in all of the eggs, the more curved end of the band being invariably in the end of the egg remote from the edge of the supporting leaf. After five days of development, the dark band has increased considerably in size and has changed somewhat in shape, showing distinct differences in the two ends, one being larger and more blunt and recognizable as the future head of the caterpillar. During the seventh to the ninth day, the black areas on either side of the head appear and the longitudinal tracheæ are visible. The

tips of the mandibles are also turning brown. At the end of the ninth or the beginning of the tenth day, the outlines of the complete caterpillar are visible through the egg capsule. It is coiled upon itself, the caudal end reflected around the head, and the dorsal surface usually turned from the surface of the leaf. The dark, chitinized head capsule and prothoracic shield are the most conspicuous parts. The intersegmental grooves are visible to a slight degree. The abdomen and the two posterior segments of the thorax are devoid of dark color or markings. At this stage, the mandibles are observed to be in active motion and contractions of the body preliminary to hatching are evident.

THE LARVA (Pl. VIII, Fig. 11).

First Instar.—Larva small; length about 1.4 mm.; maximum diameter of head 0.25 mm. Body whitish; translucent; head and prothoracic shield very dark-brown and heavily chitinized; remainder of body devoid of dark color and quite flexible. No tracheal gills. Principal setæ as in figure 11. Anal setæ 0.14 mm. long. Dorsal surface of segments often showing transverse folds and wrinkles. Prolegs with hooks arranged in transverse ellipse; usually 22–24 hooks on each body proleg; about 7–9 hooks on anal prolegs.

Case-making.—Small, elliptical cases, 2.5–3 mm. long, are constructed soon after hatching. They may be attached or independent and the leaf material may be cut from the periphery or from the middle of the leaf. The young larvæ show a very distinct tendency to remain on the lower sides of the leaves where all of the work of excising pieces of leaves is done. Cases are outgrown and new ones constructed, much as has been described for *N. maculalis*, and they seem to serve similar functions. Attached cases usually occur on the lower side of the supporting leaf.

Food.—Packard ('84, p. 825) found his supposed larva of *N. icciusalis* making cases from the leaves of *Menyanthes trifoliata*. Forbes ('10, p. 226) reported his specimens on *Potamogeton* and states that they would not eat *Limnanthemum*. Miller ('12, pp. 127, 130) found caterpillars feeding on *Potamogeton natans* and states that they may also work on *Marsilia quadrifolia*. The egg masses, larvæ, and pupæ occur predominantly on *Potamogeton natans* in the Douglas Lake region,

although a few larval cases were found, in part, composed of fragments of leaves of *Vallisneria*, a fact which suggests that this plant may be used for food. No evidence of feeding on any of the water-lilies was observed. The larvæ are active from the moment of hatching and soon begin feeding, evidence of which is the early appearance of yellowish matter in the digestive tract.

SUMMARY.

Aquatic Lepidoptera are very limited in number and the American species are little known. They rival other aquatic insects in the character of their unique habits and adaptations. Two species are considered in this paper.

I. *Nymphula maculalis* Clem.

1. Eggs are deposited only on the lower, submerged surface of the leaves of the yellow water-lily, *Nymphaea americana*, in masses arranged in concentric rows.

2. Egg masses, in the Douglas Lake region, are invariably placed about the egg holes of the chrysomelid beetle, *Donacia*.

3. Laboratory observations and experiments on females in aquaria yielded the following data: (a) Eggs were invariably deposited at night. (b) Eggs were invariably placed about *Donacia* egg holes when the latter were available. (c) Oviposition may extend over five successive nights. (d) One female may use several *Donacia* egg holes before oviposition ceases. (e) Maximum number of eggs laid by a single female was 617. (f) In the absence of *Donacia* egg holes or other similar punctures in the water-lily leaves, oviposition was usually delayed but ultimately resulted in the deposition of small egg masses on the lower side of the leaves at the margins. Egg masses were deposited about artificial punctures and incisions of various sizes and shapes, the dimensions of which apparently had little to do with the selections.

4. The egg period is about eleven days. There is a definite and constant orientation of the eggs in the mass since the heads of the larvæ invariably develop in the ends of the eggs nearest the leaf puncture.

5. The larva in the first instar is devoid of tracheal gills and respiration is apparently cutaneous.

6. Tracheal gills first appear in the second instar. Paired, lateral, filamentous gills occur on all of the segments except the prothorax and the last abdominal. Maximum number of gills per segment is two.

7. In the third instar, an increase in the number of gills appears, the maximum number being three. Dorsal, paired, filamentous gills also appear.

8. The sole method of locomotion by larvæ is an inefficient crawling. There is no evidence of an ability to swim. When supported by a case, change of position can be effected by side to side motions of a portion of the body extended into the water.

9. Case-making is a constant larval activity from time of hatching to pupation and sometimes seriously reduces the leaf surface of the food plants. The chief functions of the case seem to be (1) protection, and (2) support in the water.

10. No evidence of feeding or case construction which involved plants other than *Nymphæa americana* was observed, although the larva has been reported in connection with *Castalia* and *Brasenia*.

11. After the first instar, tracheal gills constitute the larval respiratory organs. The maximum number of gills increases with advancing age from two per segment in the second instar to as many as ten per segment in the mature larva. The total number of gills increases from twenty-two in the second instar to one hundred in the mature larva. The number of gill filaments per segment increases from four in the second instar to as high as forty-eight in the full-grown larva. The total number of gill filaments on the body of a larva increases from forty in the second instar to over four hundred in the full-grown larva.

12. Dissemination in the larval stage is accomplished by crawling over contiguous leaves; by voluntary propulsion in detached cases; by the effects of winds, waves, and currents on detached cases; and indirectly by the work of certain other insects which cause a separation of the leaves of the food plant from the petiole.

13. The pupa is described in detail. It is usually attached to the lower, submerged surface of the water-lily leaf and is enclosed in a silken covering which, in turn, is surrounded by the last larval case.

14. Pupæ are disseminated by indirect methods. Winds and waves, assisted by the work of certain insects, may scatter pupal cases widely.

15. The adult moth is largely nocturnal in habit. It suffers little, if any, from contact with the water, and in extended flights over water may drop to the surface for short rests.

II. *Nymphula icciusalis* Wlk.

16. Eggs are deposited on *Potamogeton natans*. In the aquaria, females, in the absence of *Potamogeton*, deposited eggs, after some delay, on *Nymphæa americana*.

17. Oviposition is independent of the activities of other animals.

18. Development of the egg requires ten or eleven days. As in *N. maculalis*, there is a definite and constant orientation of the egg, when deposited normally, since the head of the larva always develops in the end nearest the edge of the leaf.

19. The first instar is described for the first time.

20. Case-making, similar to that of *N. maculalis*, is a normal activity of this larva. The great majority of the cases studied were made from leaves of *Potamogeton natans* but a few were composed, in part, of fragments of leaves of *Vallisneria* which possibly is another food plant.

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EXPLANATION OF PLATES.

PLATE VII.

Figs. 1-10. *Nymphula maculalis*.

Fig. 1. Dorsal view of recently hatched larva.

Fig. 2. Dorsal view of larva in the second instar.

Fig. 3. Dorsal view of larva in the third instar.

Fig. 4. Egg as it appears immediately before hatching.

Fig. 5. Egg masses on lower surface of leaf of *Nymphæa americana*. *p*, puncture through leaf made by female *Donacia*; *d*, eggs of *Donacia* sp.; *n*, eggs of *Nymphula maculalis*.

PLATE VIII.

Fig. 6. Ventral view of pupa.

Fig. 7. Lateral view of pupa.

Figs. 8-10. Ventral view of terminal segments of pupa, showing some common forms of variation in number and arrangement of carinæ.

Fig. 11. Dorsal view of larva of *Nymphula icciusalis*.

PLATE IX.

Figs. 12-19. Leaves of *Nymphæa americana* showing characteristic work of larvæ of *Nymphula maculalis*. Figs. 12, 13, 14 and 17 show extent to which surface of food plant may be reduced. In Figs. 13, 16 and 19, the small marginal incisions are the work of young larvæ. In Figs. 12, 14 and 17, the broad marginal incisions were produced by full-grown larvæ.